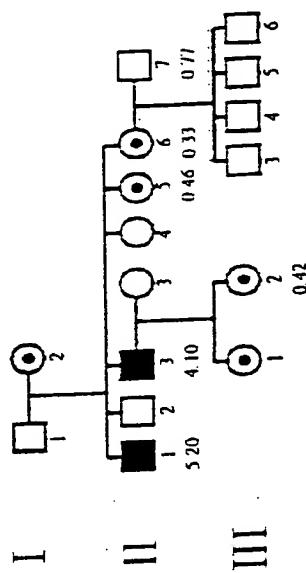
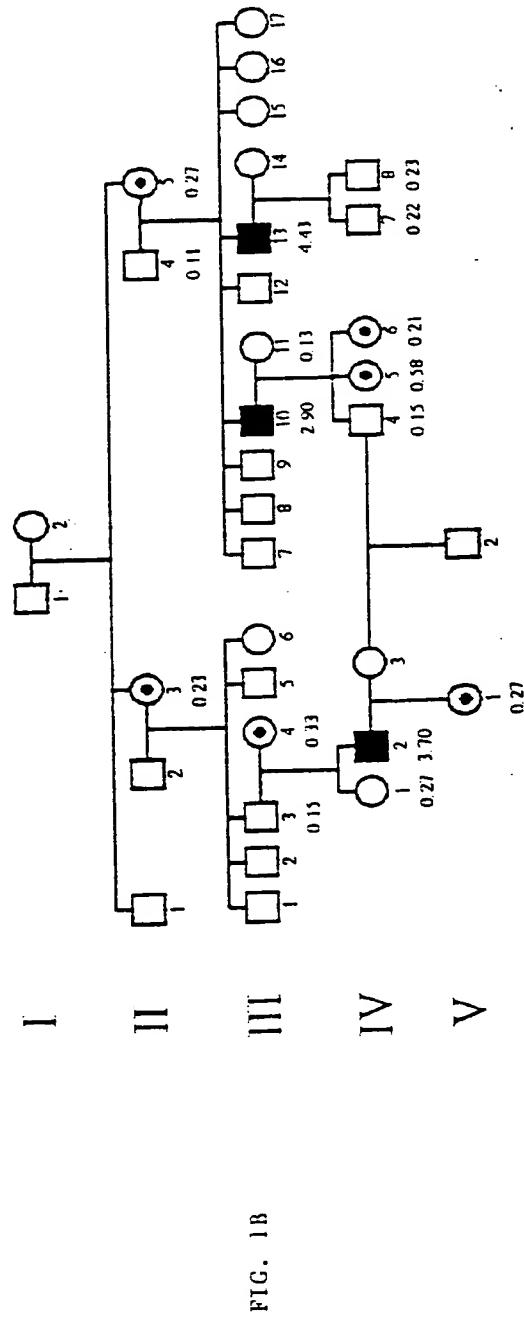


09594088-102000



096341033 - 102000

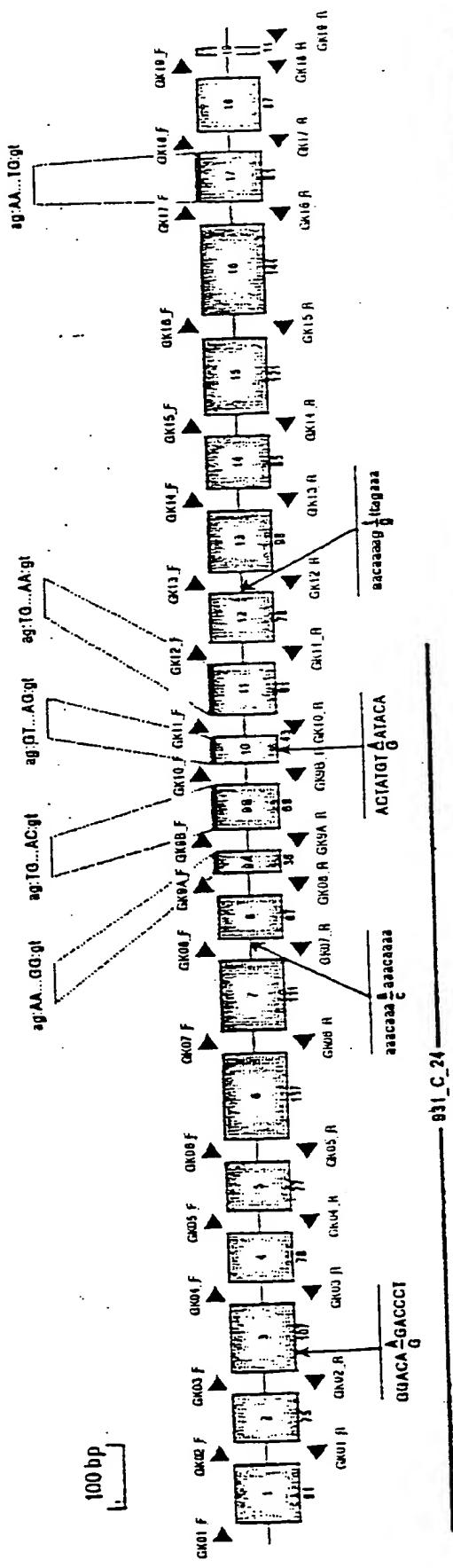


FIG. 2

Digitized by srujanika@gmail.com

DBB
N28

1

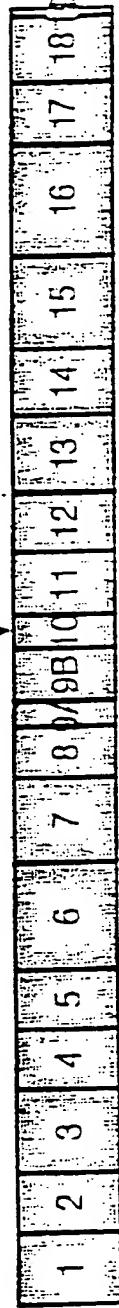


FIG. 3A

G TAT GGA ACA GGA TGT TTC TTAA CTA TGT **A** AT ACA GGC CAT AAG

GK N288D Mutant
glpk_human
glpk_rat
glpk_mouse
glpk_ecoli
glpk_pseae
glpk_entca
glpk_haelin
glpk_bacsu
glpk_yeast
glpk_mycge
glpk_enfia
glpk_mycpn
glpk_synt3

FIG. 313

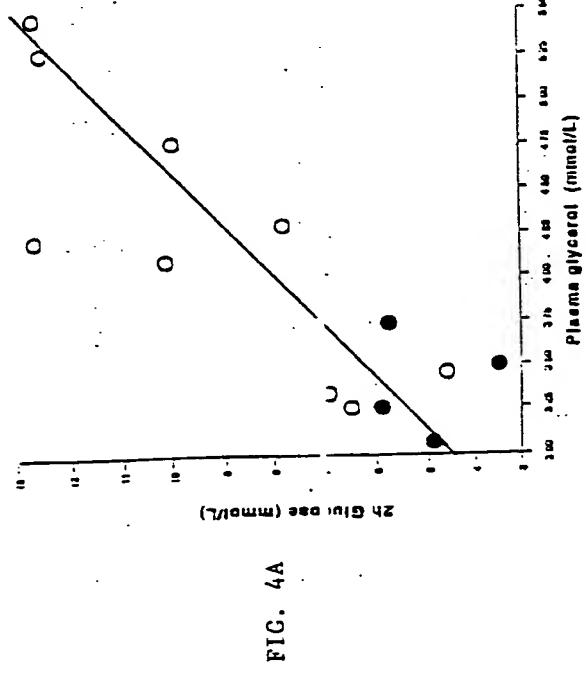


FIG. 4A

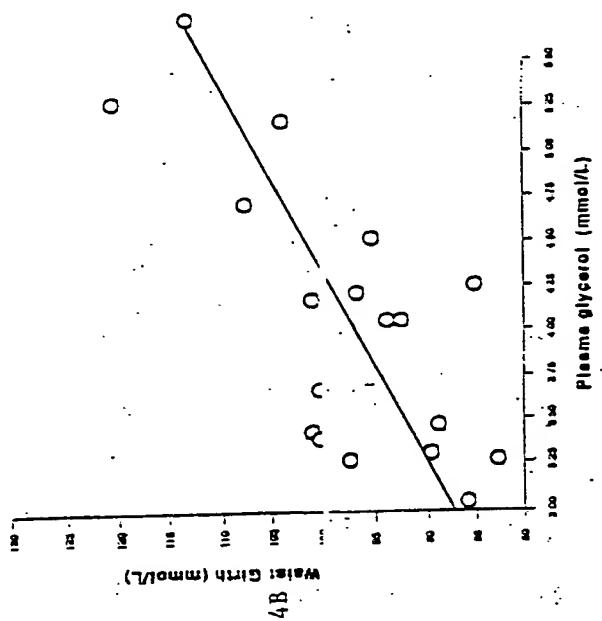


FIG. 4B

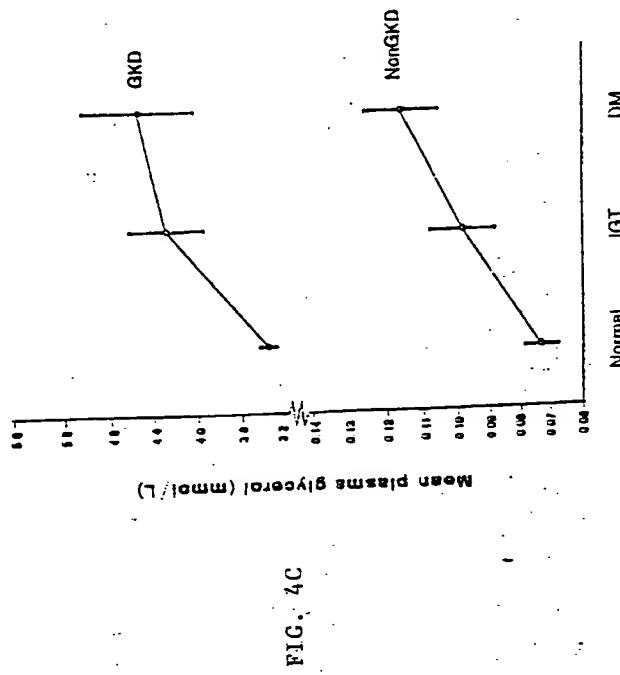


FIG. 4C

Families ranked by family mean of fasting glycerol

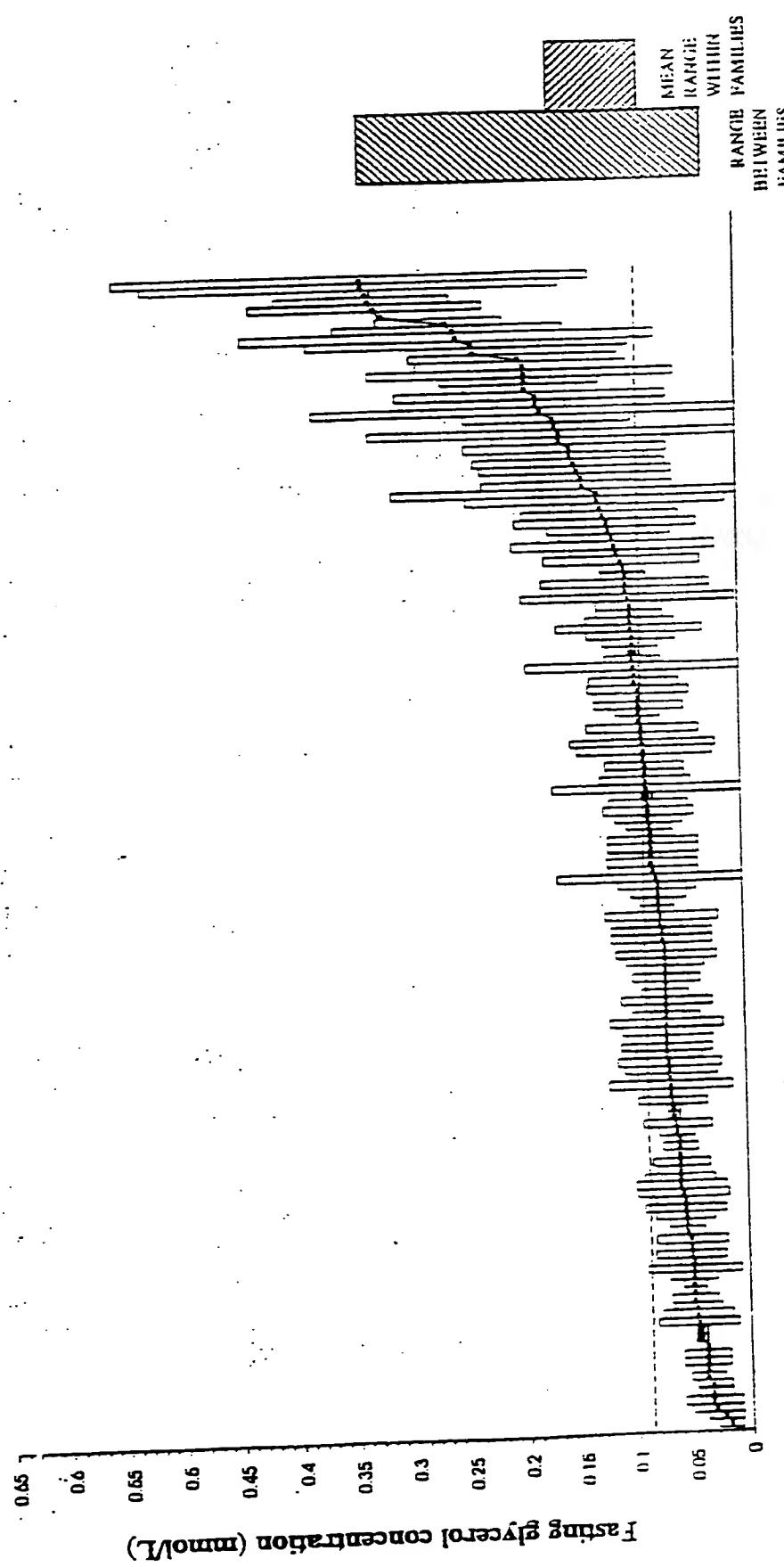


FIG. 5

poly: A/G
location: 13th base of exon 3
ATGCCTTCTTTGTCAAAGATGGGTGGAACA [A/G] GACCCTAACGGAAATTCTACAT
TCTGTCT SEQ ID NO: 1

CAA vs CAG ==> silent

poly: A/C
location: 17th base of intron 8
TAATGGTAAAAAACAAACAAA [A/C] AAACAAAAAACACACCAAAAAACCAA
SEQ ID NO: 2

poly: A/G
location: 29th base of exon 10
TTCATTCTCCCTCAACCATAAGGTATGGAACAGGATGTTCTACTATGT [A/G] AT
ACAGGCCATAAGGTtGGTTTTAATAAAAATGATTAAGTCA SEQ ID NO: 3
AAT vs GAT ==> N to D

poly: G/T
location: 22nd base of intron 12
GAAATTGGTGAGTGTGTTCTAACAAAAG [G/T] TTAGAAAATCTGAAAAATGACACA
TTTCA SEQ ID NO: 4

FIG. 6

SEQ ID NO: 5

Exon 1:
GGTCAGCGGACGGCGCGCCCTCGGTCTCTGGACTCGTACCTGCCCTCCCCCTCCCGCC
GCCGTACCCAGGAAACCGGCCGAATGCCGGCGACCTGAAGCTGGTTCATGGCAGCCT
CAAAGAAGGCAGTTGGGCCATTGGTGGGGCGGTGGACCAGGGCACCAAGTCGACGCGC
TTTTGGTAGGCCCGGGTGACATGTGAAGAGGGCGCTGAGC

Exon 2:
TGTAAAACGACGCCAGTCATCCTGATATCTGCCTGCATTACATTAATATTACAATAT
CTTTTCAGGTTCAATTCAAAAACAGCTGAACACTTAGTCATCATCAAGTAGAAATAAA
ACAAGAGTTCCAAGAGAAGGGTATGTTCTAATTAAATATGTAAAGACACATTATGTTG
TTAGTCCATCTCACCCAACTTGC

Exon 3:
CAATGCCTTCTTTGTCAAAGATGGTGGAAACA [A/G] GACCTAAGGAAATTCTACATTCT
GTCTATGACTGTATAGAGAAAACATGTGAGAAACTGGACAGCTCAATTGATATTCAA
CATAAAAGGTATTTAGTAGAATATTTACCCACA

Exon 4:
TGTAAAACGACGCCAGTTGAGAGCTGTTCTGAAGTAGTCTACTTGTAAATTGG
ACTTCCTCTGTTAACCTCTCTTAAAGCTATTGGTGTCAAGAACAGAGGGAAACCACT
GTACTCTGGACAAGATAACTGGAGAGCCTCTACAATGCTGTGGTAAGCTGTATGCAT
GGATGTCAAATGTAGGCCCTTCTCACATTGCAA

Exon 5:
TGTAAAACGACGCCAGTCCTGATAGTGTAACTTCACTAAGTTCTTAAATTGAAAG
TTTTCATGTATATTATTTATTTGGTCTATAGTGTGGCTGATCTAAGAACCCAGTCTAC
CGTTGAGAGTCTTAAAGAATTCCAGGAAATAAACTTTGTCAACGTAAGAATTCTT
CAGAACTATAACTATAAGAATGTTCTTTAAAAAGTTGCAGATTCACTAGAAAGA
AGCATTTATGGTACAATAGTTATTGATACAATTATAGAATCTTTCCCGATAATTGA
GGCC

Exon 6:
TGTAAAACGACGCCAGTTCTTTGTTGGTGGTTGTTAAACTGTTACACTTTCAT
TTGCTAATGAACTTCACAACGTCTTAACTGCTTCAAGACAGGCCTTCACTTACACTT
CACTGCACTGAAACTTCGTTGGCTCTGACAATGTGAGAAAAGTTCAAAAGGCCGTTGAAG
AAAAACGAGCTCTTTGGGACTATTGATTGATGGCTTATTGGTATGTTAAATATAATG
GATATATGGAGAATTTTCAAGAAATTCTAGACTGCCTTGCTTACTAGC
AGGTCAACTTTAATTAGCA

FIG. 7A

Exon 7:
TGTAAAAACGACGCCAGTTGTGCTCTGCTGATTATGACCCTAACATAATGTAATTAAATT
GCCAATAAGTACAAATTAAACCTGATTTTACTCTGCCTAGAGTTGACAGGAGGAGTC
ATGGAGGTGTCCACTGTACAGATGTAACAAATGCAAGTAGGACTATGCTTTCAACATTCAT
TCTTGGAATGGATAAACAACTCTGCAGTAAGTCTGTTGCTCAAATATAGTTTCC
CAATACACTACCTATTATAACCGAAATCTAATATTTAGATGTCAGTGGAGCA

Exon 8:
TGTAAAAACGACGCCAGTACAGTGTAAATACCCAATCTTCTGTTTCAGATTTTGGA
ATTCCAATGGAATTCTTCAAATGTCCGGAGTTCTGAGATCTATGCCCTAATGGTAAA
AAACAAACAAA [A/C] AAACAAAAACACACCAAAAAACCAAAACAAACAAAAAAACC
TAATAATTAAAGTTTTTATTACAAAACAAGTTACTATTCAATTCAAAGTCAACTGT
GTTATGTTGTGACTAAAAACTTACAGTCCTTTACAATGG

Exons 9A and 9B
AAAGCTGGGCCTTGGAAAGGTGTGCCAATATCTGGGTAAGTTCATCACCAAGTGTCTCCC
CATCCCCACCCTCCCCATGTTATGGCTTCCCTCTTAGTTCATCAGTGTGCCTCTTTT
AAACTAGGGAAAACAAGTAAAAGTGAAAATTGGANNNNTCTGTTCTACATGTCATACT
GTGGGCCATTGAGAATCTTTGAATAATTAAACTCTCCCTCCCATACTATTATC
TTACATATTAAACAAATGGTATTAACAAATGGGAAATGCCAAATGGAGAAAATGCAAGGA
AATAGACAGTTCAATTGATAAAATAAAAATGAAAAATAATCCTATGGCTTCTAA
AGAAAGTTAATACTATTGTATTAGTCAGTGTCTTATTGTCATTAACTTCAGTGT
GGGGACCAGTCTGTCATTGGTGGACAAATGTGCTCCAGATTGGACAAGC
GTGAGTTAACAAACAGACTAAAAACCAATGCTGTTCTTACTGGTGCTTGA
ATAAGGAAAAGCTTTGAAGTTCATCCAGGATGAAAATCAATAGCTTAATAGCTCCAATATG
CATATATACACTTTTACCATTTTATATCTTAAATAAAATACAAA
TGCCATATATATGCACACTGATGAAGCTTAAAGACCTAAATTGTAAGCTGGCGCG
GGCG

Exons 10 and 11:
TTATTTGCTTCAATAAAATTGCTTCTATTCAATTCTCCCTCAACCATAGGTATGGAACAG
GATGTTCTTACTATGT [A/G] ATACAGGCCATAAGGTTGTTTAAATTAAAAATTGA
TTTAAAGTCTAAGTTCATCTAAATAATGCTGAACATAATTACTATTAAACAAC
TCTTTAGCTTTACTTAATCTTATCAGGGTTAATTAGAGCTAACAAATTGAATC
GTTCTAATAAGAACCAATTAGACTCTTGAATTATGTTGTTTAATTGCTGG
GGGAAATCTAGACTGAGACCTCATCAAATTCTTAATGCAAATCTAATTGAAACAAGGAATA
AACTTTTATACAGCTTAAATGTGTTCTTAATTCTGATGTTGACTGTAAGGATT
TAAAAATTGGTTATTGATTGATTGATTATTGTTACCTATTGTTACTTTAACTTAA
TCTCATGTTATCTTCACTACTGAAATCTTTTTCTTACAGTGT
ATTTCAGTCACTGGCCTCTCACCAACAGTGGCTACAAACTGGCAGAGACAAAC
ATTATGCTTGGAACTAAGTTCTTAAATCAATATGGATAATATGACAAACATTCAAAGCT
AATAAAATCACAGAGTTCTAACACTTTCTGGTAAATCTTAATACAGAGACT
GTTCTGCTTCTGGCATTGATTGAGTTGAGGAACTGAAACTGATCTGGGTGT
TCACAGGAGACCTTGATTAGATTGGTCCCTCAGTCTTATGCCAATTATCATGTCAC
GGCATATTACTGAGAGCTCTACAAATGTGACGTTTTTTTATCTCTAAACTTAA
CGGATTAACGTGCTCTAACATTCTTCACTTGTGAAATTCTTGTATTATAA
TGCTCCAGTGTCCAAAGAGAACCCCTGGCACAATAGGCAGAACAACTCTTCACTGTC
TCCTCATAAAAATAATTGTGTAACATTGTATAGAAAAGAACGGAGATTATG
CCACTTATCACTGGAAACATTGTTCAAACATTGTATGTTACTAGGAATATGCCAG
CCTAACGCTATA

FIG. 7B

Exon 12:

TTTTATTAGTGA CTTAGA TAAA ACTATGTTGTATTAGAAGACCTAGTTACATATTTGT CG
GAGTCTCAAAATGGAAACTGAATTCTGTCCATCTGATTGTGTCAACACAGAAATATGCTCAA
TAAAAACCTTGGATAGTGATAAAAATATCTGTCTGAATTCTTCTTTCTTAGGGTC
TGTAGCTATAGCTGGCTGTATTCGCTGGCTAACAGAGACAATCTTGGATTATAAGACCT
CAGAAGAAAATTGGTGAGTGTGTTCAACAAAAG [G/T] TTAGAAAATCTGAAAAATGACACA
TTTCAGTATTTATCTCTGCAAAGTAAATATCGATGCTTGC CCCC AATGTGAT

Exon 13:

CCAGTTGTGTGATTTGTTTGTAAATGTTAGAAAAACTTGCTAAAGAAGTAGG
TACTTCTTATGGCTGCTACTCGTCCCAGCATTTGGCTAACATGCACCTTATTGGGAGC
CCAGCGCAAGAGGGTAAGTATTGAAAATATGGAGTGCTTGGGATCTGATTAT

Exons 14 and 15:

TGTAAAACGACGCCAGTTGATTATGTCCAATTTCTTCTGGACATTTCTGTCTACCAA
ATTTGACCTTTCATATTGAGATATTCAAATTGATTGGTTATATCATTCTAAATCTGAAA
ATCTTTGTGCGTATTTTAGGATAATCTGTGGACTCACTCAGTTCCAATAATGCCATAT
TGCTTTGCTGCATTAGAAGCTGTTCCAAACTCGAGAGGTAACAAATATGGGCTGT
TTCTTGACTTAGTCACTTTATCACTCTTAAGTTATATGTTAACACCCGAGATTTC
AGTACTGAAAATGTAGTTAACATTAAGGCTGCCAACACTAACATCTAAATATAAGCA
GGGTTTCCCCCTTTCCAGCTGTCAATTACCTCTAACGTTCTGTCCCTGTCAGGCACTG
GGAAATTATGGTGTGGGGAGGCTGAGTGGCACACATTAGGCCAACAGAACAGCACAAACA
TAGGCATCaAGGCAGAAAACAGGGTGC AAAATACAGTTGATAGCTTAGCTGAATATCAAG
GTGAATGCAGAGGTGTAGTGAGAAAAGGTTGGCTGTGACCAGATCAAAGAGGGCTTAGAA
GACCAGAATAAGAAGTCTCAATTATTCCATAGGCTCTTGAAGCTCTGAGAGTTCTGAG
TGGAGGATTGCCATTTCAGAGATGTTACTATGAAATAGATTATAACATTAAATTGCACTGG
TTTATTAAAGATTGGATGCCATGAATCGAGACTGTGGAATTCCACTCAGTCATTGCAAG
TAGATGGAGGAATGACCAGAACAAAATTCTTATGCAGCTACAAGCAGACATTCTGTATATA
CCAGTAGGTTAGTAACTCTCATTCTTAAACTCCCAGAGTAATGTTCTGTGGAATAAC
TAGTTCTTGGG

Exon 16:

TGTAAAACGACGCCAGTTCCCAGAGTAATGTTCTGTGGAATAACTAGTTCTTGGC AT
ATGTAACCACAAAGATATTGATGAACTCTCTCAGTGAAGCCCTCAATGCCGAAAC
CACTGCACTGGGTGCGGTATGGCGGCAGGGGCTGCAGAAGGAGTCGGGTATGGAGTCTCG
AACCCGAGGATTGTCTGCCGTACGATGGAGCGTTGAACCTCAGATTATGCCGAGGGT
ACATTAAAGAATGAAATGTTCACTGATAACTGTGAAAAGCACCTAGTGCACGGAGTT
TGTTTTCTGTTAGTTAAAAGTTAAGGAACCAAGTAAAATAGTAATGTTATCATTGCA
TTCGGCTGCCAACATATTGGCTTACTGAATAATGTGAATGAGAACATCGTTGCTTAT
CAAAAGAACCTCTAAACTCACTTTAAAATCATT

Exon 17:

TGTAAAACGACGCCAGTGCAGTTAATGTGTCAATAATTGTCAAGAACATGTT
GAGTGATCATAAGTATGGTACTAACAGAACATCTCAGCAAACATACCTTCTGTTATGTGTTTT
CTACCTCTAATTCTAGAAAGTGAATTCTGTTATCTACATGGAAGAACAGCTGTGATGAACT
CAATGGGTGGTTACAACATCTCCAGAAAGTGGTAAAATGTTTTGTTATTATTGT
CACATTCTTACTATATTAAATAGTTAAGTATCTAGGCATTACACATAGCCAGGCT
GCTCTGAAGAAAAGCATTATCATATGTCCAGAGATTCTGACATTGGTAAACACTTTAAAGT
TCTAAACACAAAATGTAAATTATCAGGTGT

FIG. 7C

Exon 13:
TGTAAACGACGCCAGTTGGTTGGCTTGACTGGAATCTCTCTGCTTGGATGACCA
CAGGTGACCCCTAGTATCTTCTGTAGTCTGCCCTGGGCTTTTTAGTAGTGAGTAGCATGGTA
ATGTTAACCGGAGCAAGGTACATCTCAGGTTAGTTACTCTTAAATTAGACAACCTATTAG
TTAGCTTAAATGTTTCGTGTATAACTTAGCAGAAAATTTCACTGTTTCATTCTTCTG
TGTCTAGGAAGCTGGAAAATCAATTAAAGGTCTAATTAGTTAGACCAATTAAATCTTGGGGG
CAGTTAGAAGTAAGAACTGTGACTCTGCTTACCCCTTTAAATTAAATGTGATGACTTCT
TTAAGAGGGACTACATTCTGCTGTCAGCTGCAGCAATAAGCAAAAGTGAAGAAATACTAATATT
TAAATGACAGGACTTCTAGACTGCTGAAAGTTAAAGTATACTT

Exon 19 :
AAAATTACTGGCTTAAATGGAAATGATGCTTCTTATTCTGTATGTTCCCATGAAAAGTGAAAC
TTAAAAAAAAATTCACTGATTAGGGTTCATGAAAAGGCCTGTTCTATGAAAATTGAGAC
AGGTTGCATCTCTAAGCTAAAAGATGGCTATGTGTCTAGACTCTAGACTCTAAAAATG
CATGTGGTCACTATATGTAGGTTATCTCTCGGTGACATACACTGCAATTGAGAGGGCTGG
AAATTGTTGCCTTGGTAAACGATTAGCAACAGTGGCAATTGTTAATTGGAAATTGGC
CCTGTTGTCATTAAATTGTGAGGCATGATTAGAAATCATATGGACTTCTAGCTTAA
TAAATGATTGAATCATCTGCATTGTTAACTCCTGAATTGTATGCATGTATTATTGACATA
TATGGTTTGTCCCCATTCAAGGTATTCCATAAAACCTACCAACTCATGGATTCCAAGA
TGTGAGCTTTTACATAATGAAACAACCAGCAATTCTGCTCTTAATGCAATGACACTATT
CATAGACTTTGATTTATTATAAGCCACTGCTGCATGACCCCTCAAGTAGACCTGTGGCT
TAAATAAAGAAAATGCAGCAAAAGAATGCTATAGAAATATTGGTGGTTTTTTTTTT
TAAACATCCACAGTTAAGGTTGGGCCAGCTACCTTGGGCTGACCCCTCCATTGCCATAA
CATCCTGCTCCATTCCCTCAAGATGTAGGAAGAATTGGATCCTTACCAATTGGAATCTTCC
ATCGAACATACTCAAACACTTTGGACCCAGGATTGAGTCTCTGCATGACATATACTTGATT
AAAAGGTTATTACTAACCTGTTAAAATCAGCAGCTTTGCTTTAAGAGACACCCCTAAAA
GTCTTCTTCTACATAGTTGAAGACAGCAACATCTTCACTGAATGTTGAATAGAAACCTC
TACTAAATTATAAAATAGACATTAGTGTCTCACAGCTGGATATTCTGAAAAGTTA
TTTGCACAAACTGAAATCCTCAGATGTTCCATGGTCCCCTAATTATAATGACTTCTG
TCTGGGTCTTATAGGAAAAGATACTTTCTTTTCTTCATCTTCCATTCTTCTTTATATT
CTTGTATGTATAACATACATGCCTATATATTATACACTGAGGGAGCCATTATAAATA
AAGACCACATATATTCAAGAAGGTTCTAACAGGG

DODD 04060801 01020200

TABLE 1

Characteristics of carriers of the N288D GK gene mutation
and of their unaffected relatives

	Men			Women		
	N288D carriers	Unaffected relatives	p	N288D carriers	Unaffected relatives	p
N	18	18		14	14	
Age (years)	46.4±14.2	42.0±18.8	0.32	44.9±13.5	43.7±17.8	0.37
Uncorrected triglyceride (mmol/L) ⁽¹⁾	6.26±1.13	2.05±0.54	<0.0001	2.84±1.20	1.30±0.65	0.0002
Glycerol (mmol/L)	3.99±0.71	0.10±0.04	<0.0001	0.54±0.14	0.10±0.02	<0.0001
Corrected triglyceride (mmol/L) ⁽¹⁾	2.27±0.75	1.95±0.53	<0.0001	2.31±1.22	1.19±0.67	0.03
Free fatty acid (mmol/L)	0.77±0.22	0.57±0.25	0.01	1.29±0.35	0.76±0.17	0.0004
Fasting glucose (mmol/L)	5.2±0.74	4.8±0.31	0.13	5.0±0.7	4.6±0.3	0.10
2h glucose following OGTT (mmol/L)	7.9±3.1	5.8±1.6	0.02	7.0±6.1	5.0±2.1	0.04
Fasting insulin (mU/L) ⁽¹⁾	13.3±14.0	15.1±14.8	0.62	12.2±13.1	9.0±3.4	0.60
Waist girth (cm)	97.7±9.3	88.1±12.3	0.01	88.5±5.3	79.3±5.3	0.03
Body mass index (kg/m ²)	27.9±4.1	24.9±3.9	0.03	28.1±5.5	23.1±7.3	0.001
% total body fat	27.1±7.2	22.9±7.6	0.01	46.3±3.1	33.9±11.3	0.001

⁽¹⁾ geometric mean, p value after log transformation

TABLE 2

Fasting plasma glycerol concentration (mmol/L) in the initial cohort of 1056 individuals,
by risk factor of glucose intolerance and diabetes mellitus

		No.	Glycerol geometric mean \pm SD	p
Gender				
	men	717	0.065 \pm 0.081	-
	women	- premenopausal	0.071 \pm 0.093	<0.0001
		- menopausal	0.099 \pm 0.085	
Age (Y)				
	<40	486	0.071 \pm 0.082	-
	50 - 60	408	0.076 \pm 0.106	0.0015
	>60	165	0.083 \pm 0.053	
Fasting glucose (mmol/L)				
	<5.7	449	0.068 \pm 0.080	-
	5.7 - 5.9	336	0.070 \pm 0.090	<0.0001
	6.0 - 6.9	271	0.090 \pm 0.100	
Fasting insulin (U/L)				
	<15	637	0.067 \pm 0.082	0.02
	≥ 15	419	0.086 \pm 0.101	
2 hours glucose (mmol/L)				
	<7.3	572	0.062 \pm 0.071	-
	7.3 - 11.0	283	0.081 \pm 0.101	<0.0001
	≥ 11.1	201	0.102 \pm 0.110	
Triglyceride (mmol/L)				
	≤ 2.2	389	0.057 \pm 0.062	<0.0001
	>2.2	667	0.082 \pm 0.103	
Free fatty acid (mmol/L)				
	<0.6	589	0.066 \pm 0.054	<0.0001
	≥ 0.6	467	0.111 \pm 0.112	
Body mass index (kg/m ²)				
	≤ 27	428	0.060 \pm 0.087	<0.0001
	>27	628	0.079 \pm 0.097	

p value from one-way ANOVA

Figure 9

TABLE 3. Multivariate analysis of the relationships of fasting plasma glycerol concentration with impaired glucose tolerance (2h glucose 7.8-11.0 mmol/L following a 75 g oral load) before and after adjustment for covariates identified in

	Model 1	Model 2	Model 3	Model 4
Glycerol (log)				
B	1.75	1.62	1.46	0.7
Odds ratio	5.76	5.42	4.33	2.4
p	<0.0001	<0.0001	<0.0001	0.0
Triglyceride (log)				
B	0.54	0.35	0.18	
Odds ratio	1.75	1.42	1.11	
p	0.02	0.11	0.63	
Body mass index (kg/m ²)				
B		0.10	0.05	
Odds ratio		1.10	1.05	
p		<0.0001	0.01	
Fasting insulin (log)				0.57
B				1.31
Odds ratio				0.39
p				
Fasting glucose (mmol/L)				1.13
B				2.65
Odds ratio				<0.0001
p				
Free fatty acid (log)				1.62
B				4.13
Odds ratio				0.07
p				

Odds ratios are expressed as the increase in the risk of 2h glucose >7.3 mmol/L following a 75 g oral charge, associated with a 1-SD increase in the variables studied. B denotes the standardized estimate which is the parameter estimate of each variable in the multivariate logistic model. All models included age and gender as covariates. Otherwise, only the variables included in each model are shown. Subjects with severe hyperglycemia due to the N788D mutation in the

Figure 10